

## **METHOD FOR ASSAYING MULTI-COMPONENT MIXTURES**

### **FIELD OF INVENTION**

[0001] The present invention relates to analysis of substances and materials by optical methods, particularly for identifying and assaying multicomponent mixtures, as well as for analytical screening of trace amounts of contaminants in complex media, including potentially anthropogenic contaminants in objects of life support and the environment

### **BACKGROUND OF THE INVENTION**

[0002] Practically all things, both natural and man-made, in any aggregative state, are multi-component mixtures. Even super-pure chemically individual substances that have been subjected to multiple purification steps always contain a set of trace impurities. For a number of reasons, it is nearly impossible, using current techniques, to separate these impurities from the desired component.

[0003] For example, even ideally purified deionized water contains trace amounts, at least, of dozens of humus impurities (Lavrik et al., 2000). Distilled spirits produced by the highest separation efficiency contain more than 200 trace components (Karagodin, 1998). Marketable cyclohexane contains up to 70 trace components (Dedkov, 2002). Yet these three entities are considered to be pure substances.

[0004] The situation with industrial products is much more complicated, because in making these products a certain set of impurity components are introduced as part of the process of production, and these impurities define the quality and consumption features of a product. Other components, which are sometimes present in trace amounts, define the safety of the product.

[0005] At present, about 80,000 kinds of chemical products are manufactured all over the world, most of which products are toxic to warm-blooded animals (Korte, 1996). The appearance of these substances in the environment and, particularly, in life support systems, poses a serious threat. The problem of water quality on the planet is particularly important, because 90% of the water is consumed for industrial use and is returned to the environment with wastewater (Dedkov, 2002). In fact, more than 500 individual man-made substances have been detected in wastewater, almost half of which are regarded as very dangerous, while the rest are characterized as dangerously harmful to vital functions and the functioning of ecosystems (Fortoutan-Red, 1982).

[0006] In other words, in practically all fields of human activities, there is a need for effective analytical techniques so that dangerous substances can be detected and eliminated.

[0007] Traditional methods for analyzing compounds in multi-component mixtures are usually based on extraction of the analyte from the mixture, with further qualitative-quantitative correlation. This procedure requires a number of instrumental physical and chemical methods, often resulting in the requirement for preliminary treatment or concentration of a sample. Modern standardized analytical processes cover only about 20% of the total available set of components (Devjatykh et al., 1994). This results from the fact that the substances being analyzed may consist of tens and hundreds of individual compounds, and exhaustive analysis of the all of the components of a multi-component mixture becomes a very long, laborious, and expensive procedure. At the same time, modern conditions dictate the requirement for rapid analysis methods which are simple in execution and which are able to rapidly detect the widest array of contaminants.

[0008] The methodology of analytical screening (Beyermann, 1982) relates to such methods, which itself is not strictly an analytical procedure. The essence of this methodology consists in rapid screening of suspicious objects requiring further standard by stricter methods. The parameters of these objects deviate from the parameters of standard object by any characteristic which is subject to rapid methods of instrumental control. It is evident that, for multi-component mix-

tures, this characteristic is the identity or distinction of a compound in the product being analyzed, as compared with a standard compound which corresponds strictly with the manufacturer's process, or with a natural set of natural components wherein absence of the contaminants is confirmed. Thus, the solution of the task of rapid analysis does not require decoding of a qualitative-quantitative compound of a mixture. For that, it is quite enough to find an analytic method making it possible to receive some integral characteristic, or a set of characteristics, which can be measured instrumentally, ensuring unique identification of a compound in a mixture, and, at the same time, making it possible to detect qualitative changes of the compound at the level of trace amounts of a substance.

[0009] Optical spectrum methods of analysis are the most attractive for analyzing multi-component mixtures. One peculiarity of optical spectra is the fact that they are characterized by complex reflection of both the component compound of the mixture and characteristics of individual components related to their chemical structure, and a complicated set of interactions of the components of the mixture with each other of a non-covalent nature (supra-molecular interactions [Lenn, 1995]. These interactions define the per-molecular structure of a substance of nano-dimensional scale that is responsible

for producing the optical spectra (Bakhshiev, 1972; Lakowicz, 1983; Suppan, 1990). In view of these circumstances, the optical spectra of multi-component mixtures are uniquely sensitive to the slightest changes of a component in the mixture, which spectra appear both owing to mechanisms of intramolecular photonics and as a result of the influence of supramolecular interactions upon spectroscopic visualization of these mechanisms.

[0010] Photo-absorption methods are the most widely used analytical techniques. These methods are based on relative measurement of the light falling on a sample and passing through the sample. When these measurements are performed, systematic hardware inaccuracies are taken into account automatically, making these methods suitable for quantitative assessments. So, in particular, methods are known to analyze liquids for determining their identity or differences. Likewise, methods are known for determining contaminants in liquids, which methods consist of measuring absorption or reflectance spectra of an unknown liquid to determine characteristic spectra, finger prints, and/or profiles of data, and to determine the ratio of intensities of the light reflected or absorbed by a standard sample and a tested sample within the selected spectral section (Ingrum, 1991; Littlejohn, 1991). However, the absorption and reflectance spectra have low sen-

sitivity and are ineffective for identifying trace amounts of impurities.

[0011] The luminescence spectra (Siegel et al., 1985) are much more responsive to trace amounts of impurities and are very selective. At the same time, for identifying components of a mixture, three dimensional luminescence spectra (Webor, 1961) are most appropriate, which spectra reflect the complete set of spectral-luminescent characteristics of the test object. These spectra may be represented both as an isometric projection in coordinates of excitation wave length and irradiation wave length, or contoured spectra, as shown in Figures 1 and 2. Particularly, there is a known method for identifying a petroleum type by contoured luminescence excitation-irradiation spectra (Rho et al., 1978; Corfield et al., 1981). Also, very striking differences are observed for petroleum synthesis products. An illustration of this is found in Figure 1 in which the contoured spectra for various brands of gasoline are shown.

[0012] The drawbacks of the luminescence spectra are that the excitation-irradiation spectra are not able to display a complete component makeup of a sample, but only part of the components. To receive contoured spectra, mathematical interpolation software is used, which inevitably distorts the analysis result, making them of little use for correct identi-

fication and/or determination of differences for mixtures which are similar to each other. More, as stated in Siegel et al., 1985, visual analysis by three-dimensional spectra for mixtures containing more than three components presents a quite insoluble problem. For example, Figure 2 shows the contoured spectra for luminescence of tap water and the same water with added chlorine. As one can see, the visual differences of contoured spectra for these samples are imperceptible.

[0013] The closest solution has been the method of identification of spirit-based liquids by the difference in matrices composed using a complex set of spectral profiles of a light transmission ratio, and luminescence profiles normalized for a unit, when excited by different wavelengths of light (Nekrasov, 2000). This method totally excludes the human factor, resulting in positive identity and differences in trace amounts of impurities, even for objects which are very close in their composition (Nekrasov et al., 2000). The drawbacks of this method are that analytical matrices used for identification contain data of absolute measurements of the intensities being analyzed, and do not take into account the spectral light transfer ratio of the optical path of an analytic device, and the spectral responsiveness of photo-detectors. This makes it impossible to correctly compare the analysis re-

sults and practically excludes their use in forming centralized electronic data banks. Also, during the actual measurement, noise caused by intensity fluctuations of the light source, being particularly perceptible in the ultraviolet range of the spectrum, is not taken into account, thus influencing the selectivity of the method and its possibilities for identification.

[0014] At the same time, the spectral characteristics used in the methods described above (Corfield et al., 1981; Siegel et al., 1985; Nekrasov, 2000) reflect, mainly, only those components of a mixture that have pronounced chromophore characteristics and/or luminescence. In both absorption and luminescence of multi-component systems, the components not having their own chromophore and luminophore characteristics are shown weakly. Both methods, by objective reasons have an analytical range which is restricted by the high concentrations of components of the mixture, resulting in drastic restriction of the possibilities of identification where there are high concentrations of characteristic components. These circumstances do not allow one, particularly for ultra-clean substances and compounds, to use referencing of the objects into classes and to effectively use on-line features of communications networks and electronic data banks for identification.



It makes it impossible to identify unknown objects in the physical absence of a standard sample of a compound.

#### **SUMMARY OF THE INVENTION**

[0015] It is an object of the present invention to overcome the aforesaid deficiencies in the prior art.

[0016] It is an object of the present invention to provide a method for determining the identity of components in multi-component mixtures.

[0017] It is another object of the present invention to detect contaminants in multi-component mixtures.

[0018] According to the present invention, a highly effective method is provided to determine the identity or differences between multi-component mixtures, as well as to detect contaminants in multi-component mixtures, by means of comparing the spectroscopic parameters set, which parameters completely characterize a component in the mixture.

[0019] The present invention provides a reagent-free express instrumental method for registering and analyzing electronic optical signatures of multi-component mixtures. This method is suitable for forming dynamic computer-based data banks, making it possible to control the content of industrial products, life support systems, and the environment, as well as to prevent harmful and dangerous contaminations in these systems.

[0020] According to the present invention, the complex responsiveness of objects which are analyzed to the light influence by means of comparing absorption, spectral, luminescent, and Raman characteristics of the object being analyzed. These characteristics were obtained while performing relative measurements of characteristics of compounds being identified and characteristics of a standard sample and/or a calibration sample of a known compound by a quasi-doublebeam method ensuring calculation and compensation of systematic and random hardware inaccuracies directly during implementation of comparative measurements and/or with further mathematical treatment of the results.

[0021] A complex analysis using three complementary methods, photo absorption, luminescence, and Raman scattering, promotes increasing responsiveness and selectivity of the method (Beyermann, 1982). The high express degree of analysis is secured by obtaining the complete set of absorption, spectral and luminescent, and Raman characteristics during the united analytical cycle of measuring and comparing the characteristics of samples under comparison in each of the selected points of the specific analytical range in one specialized analytical device, and/or while comparing the measured characteristics of a sample being identified in an unknown mixture

with the electronic signature of a standard sample of a known mixture.

[0022] Measurements of transmitted or reflected light are executed using standard methods, such as those shown in Weber, 1961 and Rabek, 1982. Intensity of luminescent radiation irradiated by a sample is measured in accordance with methods described by Lakowicz, 1983; Parker, 1968; and Rabek, 1982. At the same time, measurements are performed for intensity values of the exciting light  $I_0$  at the excitation wavelength  $\lambda_i$ , and the intensity values of signals being measured are normalized for this value. Calculation and compensation of the light transfer ratio for an optical path of the registering and measuring apparatus are performed by means of the procedure of relative measurements of intensity values for the sample being identified and a standard sample and/or specially prepared calibration sample of luminescent mixtures of a certain, strictly unchangeable compound with stable by time spectral and luminescent characteristics.

#### **BRIEF DESCRIPTION OF THE DRAWINGS**

[0023] FIG. 1 shows contoured luminescence excitation-irradiation spectra of motor gasoline of different brands. Figure 1 A shows the spectra for A-76. Figure 1 B shows spectra for A-93.

[0024] FIG. 2 A shows contoured luminescence excitation-irradiation spectra of clean water. Figure 2 B shows the same water containing an impurity of active chlorine.

[0025] FIG. 3 is a block diagram of a specialized analytical device (a) and diagram of switching positions of samples when measuring the intensity values of the probing light  $I_0$  (b), the passing light  $I_T$ , and the luminescent light  $I_L$  (or the Raman scattering  $I_R$ ) for a standard sample of a known mixture  $K$  (c) and a sample being identified of an unknown compound  $U$  (d):

LS - a light source,

$\lambda_1$  SM - a device for extracting the monochromatic probing irradiation,

$F_1$  - a device of focusing the probing ray;

SH - a holder of samples with a device for replacing a standard sample and a sample being identified in the analytical channel:  $K$  is a standard sample of a known mixture,  $U$  is a sample being identified of an unknown compound;

$F_{2,3}$  - devices for gathering the probing light  $I_0$ , the passing light  $I_T$ , and the luminescent light  $I_L$  (or the Raman scattering  $I_R$ ) to photo-detector devices,

$\lambda_j$  SM - a device for extracting the luminescence wavelengths (or the Raman scattering),

PD<sub>1</sub>, PD<sub>2</sub> - photo-detector devices for measuring the intensity

values, accordingly, for the probing light and the passed light, and the luminescent light or the Raman scattering; R&RM - a module of registration and treatment of signals including power and control units (remote control) of execution units, and a module of communication with an external computing device and/or a device for displaying the analysis results.

[0026] FIG. 4 shows spectra of intensity ratios for impurity luminescence of water samples with the same fixed excitation wavelength ( $\lambda_{\text{ex}} = 230 \text{ nm}$ ):

- centralized municipal water supply system (curves 1-3);
- the same water contaminated with municipal wastewater (curve 4);
- the same water contaminated with 3,4-benzopyrene (curve 5);
- the same water contaminated with petroleum derivatives (curve 6).

#### **DETAILED DESCRIPTION OF THE INVENTION**

[0027] The present invention can be described by the following non-limiting examples, which examples are given for illustration only and not for limitation.

##### **Example 1**

[0028] a. Light from a wide-band source of optical radiation is divided into spectral components, and/or the assigned

sections of monochromatic light with a spectral width of  $\Delta_i$  are extracted from this light in the selected range of wavelengths  $\lambda_i$  while ensuring the possibility of changing the spectral position of these sections with the assigned pitch  $\Delta\lambda_i \geq \Delta_i$ . The extracted monochromatic light is then focused onto a probing beam having a specified geometric shape, and the light is directed onto a sample under analysis;

[0029] b. A sample of a known mixture  $K$  and a sample of an unknown mixture  $U$ , are irradiated, by turns, with monochromatic light at the assigned wave-lengths  $\lambda_1, \lambda_2, \dots \lambda_m$  from the selected spectral range  $\lambda_i$ ;

[0030] c. Light falling onto a sample, passing through a sample, or reflected by a sample, and the luminescent light is collected and directed to independent photo-detector devices;

[0031] d. The intensity values of the light gathered on the photo detector is measured for each of the extracted wave-lengths  $\lambda_1, \lambda_2, \dots \lambda_m$  of the selected spectral range  $\lambda_i$ . That is, the probing light  $I_0$ , the light passing through a sample or reflected by a sample  $I_T$ , and the luminescence light of a sample  $I_L$ , including steps of decomposition of the luminescent light into a variety of spectral sections, and/or sequential extraction of the specified sections of wave-lengths of  $\lambda_1, \lambda_2, \dots \lambda_n$  with a width of  $\Delta_j$ , with the assigned pitch of  $\Delta\lambda_j \geq \Delta_j$ ,

where each section corresponds with the predetermined characteristic wave-length of the light in the selected spectral range  $\lambda_j$ , which is gathered and registered by a photo-detector is measured.

[0032] e. The relative intensity of the gathered light passing through a sample, or reflected by a sample, is defined at each of extracted wave-lengths  $\lambda_m$ , and the luminescent light for each of the extracted wave-lengths  $\lambda_m$ ,  $\lambda_n$  for a known mixture and unknown mixture, in accordance with the following expressions:

$$T^K(\lambda_m) = \frac{I_T^K(\lambda_m)}{I_0^K(\lambda_m)}; \quad T^U(\lambda_m) = \frac{I_T^U(\lambda_m)}{I_0^U(\lambda_m)};$$

$$L^K(\lambda_m, \lambda_n) = \frac{I_L^K(\lambda_m, \lambda_n)}{I_0^K(\lambda_m)}; \quad L^U(\lambda_m, \lambda_n) = \frac{I_L^U(\lambda_m, \lambda_n)}{I_0^U(\lambda_m)}, \text{ where}$$

$I_T^K(\lambda_m), I_T^U(\lambda_m)$  - intensity values of passing or reflected light for samples of a known  $K$  mixture and an unknown  $U$  mixture measured within the specified sections  $\lambda_1, \lambda_2, \dots, \lambda_m$  of the extracted range of wave-lengths of incident radiation  $\lambda_1$ ;

$I_L^K(\lambda_m, \lambda_n), I_L^U(\lambda_m, \lambda_n)$  - intensity values of the luminescent irradiated by a sample of a known  $K$  mixture and a sample of an unknown  $U$  mixture within the specified sections  $\lambda_1, \lambda_2, \dots, \lambda_n$  of the extracted range of wave-lengths  $\lambda_j$  with light excitation within the specified sections  $\lambda_1, \lambda_2, \dots, \lambda_m$  of the extracted

range of wave-lengths  $\lambda_i$ ;

$I_0^K(\lambda_m), I_0^U(\lambda_m)$  - intensity values of incident probing radiation falling on the appropriate samples at the time of performing measurements;

[0033] f. The corresponding relative intensity values of the light passing through a sample, or reflected by a sample, on each of extracted wave-lengths  $\lambda_m$ , and the luminescent light for each of the extracted wave-lengths  $\lambda_m, \lambda_n$  for a known mixture and an unknown mixture are compared, and the compliance between the unknown mixture and the known mixture is defined by the following expression:

$$A = \frac{1}{2} \times \left( \frac{1}{m} \times \sum_{m=1}^m \frac{T^U(\lambda_m)}{T^K(\lambda_m)} + \frac{1}{m \times n} \times \sum_{m=1}^m \sum_{n=1}^n \frac{L^U(\lambda_m, \lambda_n)}{L^K(\lambda_m, \lambda_n)} \right) = 1 \pm \delta, \text{ where}$$

$T^K(\lambda_m), T^U(\lambda_m)$  - intensity values of passing or reflected light for samples of a known  $K$  mixture and unknown  $U$  mixture, normalized for the intensity of incident radiation falling on a sample at the corresponding wave-length;

$L^K(\lambda_m, \lambda_n), L^U(\lambda_m, \lambda_n)$  - intensity values of the luminescent light irradiated by a sample of a known  $K$  mixture and unknown  $U$  mixture within the specified sections  $\lambda_1, \lambda_2, \dots, \lambda_n$  of the extracted range of wave-lengths  $\lambda_j$  with light excitation within the specified sections  $\lambda_1, \lambda_2, \dots, \lambda_m$  of the extracted range of wave-lengths  $\lambda_i$ , normalized for intensity of incident radiation



falling on a sample at the corresponding wavelength;

m, n - a number of extracted sections with the wave-lengths under compared within the selected spectral ranges  $\lambda_i$  and  $\lambda_j$ , accordingly;

$\delta$  - a value of allowable deflections of the compared values for the unknown mixture being identified from the corresponding values for the similar values of a standard sample of a known mixture;

[0034] g. The presence or absence of foreign impurities (contaminants) in the mixture under study is defined by the following expressions:

$$C_T(\lambda_i) = \frac{T^U(\lambda_{m+1})}{T^K(\lambda_{m+1})} - \frac{T^U(\lambda_m)}{T^K(\lambda_m)}; \quad C_L(\lambda_i, \lambda_j) = \frac{L^U(\lambda_{m+1}, \lambda_{n+1})}{L^K(\lambda_{m+1}, \lambda_{n+1})} - \frac{L^U(\lambda_m, \lambda_n)}{L^K(\lambda_m, \lambda_n)};$$

at the same time, if  $C_T(\lambda_i) = C_L(\lambda_i, \lambda_j) = 0 \pm \delta$ , foreign impurities in the sample being identified are absent, and the conclusion about the presence of unwanted contaminations in the mixture under study may be drawn when there are sections  $C_T(\lambda_i) > 0 \pm \delta$ , and/or  $C_L(\lambda_i, \lambda_j) \neq 0 \pm \delta$  in the difference spectra.

## Example 2

[0035] In another embodiment of the present invention, the procedure differs from the procedure of Example 1 as follows: the step of comparing an unknown sample with a known one is replaced with the step of comparing their electronic ab-

sorption-luminescent signatures (ALS), for which purpose the following steps are to be added:

[0036] a. Calibration samples  $Q$  are prepared, which calibration samples represent a specially made-up mixture or multi-component solutions of a strictly identical constant compound, the absorption or reflectance spectra, and luminescence spectra of which solution (mixture) are overlapped with the corresponding spectra of a known mixture within the selected analytical spectral ranges  $\lambda_i$  and  $\lambda_j$ , i.e. have nonzero intensity in the specified ranges;

[0037] b. A standard sample of a known  $K$  mixture and a calibration  $Q$  sample are placed into an analytical device;

[0038] c. Measurements and transformations of measured values for intensity of the light passing through or reflected by standard and calibration samples, and their luminescent light, are performed in accordance with steps a-e of Example 1, while taking into account the step of replacing an unknown  $U$  sample with the calibration  $Q$  sample;

[0039] d. An electronic absorption-luminescent signature  $ALS_K$  of a standard sample is created as follows:

$$ALS_K = \|K_E\| = \left\| \begin{pmatrix} \frac{T_1^K}{T_1^Q} \end{pmatrix} \begin{pmatrix} \frac{L_{1,1}^K}{L_{1,1}^Q} \end{pmatrix} \dots \begin{pmatrix} \frac{L_{1,n}^K}{L_{1,n}^Q} \end{pmatrix} \right\|, \text{ where:}$$

$$\left\| \begin{pmatrix} \frac{T_m^K}{T_m^Q} \end{pmatrix} \begin{pmatrix} \frac{L_{m,1}^K}{L_{m,1}^Q} \end{pmatrix} \dots \begin{pmatrix} \frac{L_{m,n}^K}{L_{m,n}^Q} \end{pmatrix} \right\|$$

$T^K(\lambda_m), T^Q(\lambda_m)$  - intensity values of passing or reflected light for a sample of a known  $K$  mixture and a sample of a calibration  $Q$  mixture, normalized for intensity of incident radiation falling on a sample at the corresponding wave-length;

$L^K(\lambda_m, \lambda_n), L^Q(\lambda_m, \lambda_n)$  - intensity values of the luminescent light irradiated by a sample of a known  $K$  mixture and a calibration  $Q$  mixture within the specified sections  $\lambda_1, \lambda_2, \dots, \lambda_n$  of the extracted range of wave-lengths  $\lambda_j$  with light excitation within the specified sections  $\lambda_1, \lambda_2, \dots, \lambda_m$  of the extracted range of wave-lengths  $\lambda_i$ , normalized for intensity of incident radiation falling on a sample at the corresponding wave-length;

[0040] e. An electronic signature of a standard sample  $ALS_K$  is entered into the computer database, and/or is saved on an intermediate medium;

[0041] f. Steps similar to those in step c of this embodiment are carried out with a sample of an unknown mixture, including the step of replacing a standard sample  $K$  with a calibration sample  $Q$ , which is strictly identical with the calibration sample used when measuring a standard sample, and creating an electronic signature  $ALS_U$  of a sample being identified:

$$ALS_U = \|U_E\| = \left\| \begin{pmatrix} \frac{T_1^U}{T_1^Q} \end{pmatrix} \begin{pmatrix} \frac{L_{1,1}^U}{L_{1,1}^Q} \end{pmatrix} \dots \begin{pmatrix} \frac{L_{1,n}^U}{L_{1,n}^Q} \end{pmatrix} \right\|;$$

$$\left\| \begin{pmatrix} \frac{\ddot{T}_m^U}{T_m^Q} \end{pmatrix} \begin{pmatrix} \frac{\ddot{L}_{m,1}^U}{L_{m,1}^Q} \end{pmatrix} \dots \begin{pmatrix} \frac{\ddot{L}_{m,n}^U}{L_{m,n}^Q} \end{pmatrix} \right\|;$$

[0042] g. Comparing electronic signatures of an unknown mixture  $ALS_U = \|U_E\|$  being tested with similar data for a standard sample of a known mixture  $ALS_K = \|K_E\|$  by the following expression:

$$\|A_{ALS}\| = \frac{\|U_E\|}{\|K_E\|},$$

at the same time, a conclusion on the identity of the unknown mixture and the certain mixture may be drawn when all cells of the matrix  $\|A_{ALS}\|$  contain only unit elements (i.e.  $a_i = a_{i,j} = 1 \pm \delta$ );

[0043] h. To define the presence or absence of unwanted contaminants for the mixture under study by the following expression:

$$\|C_{ALS}\| = \frac{\|U_E\| - \|K_E\|}{\|K_E\|},$$

[0044] at the same time, a conclusion about the presence of unwanted contaminations in the mixture  $\|C_{ALS}\|$  under studying may be drawn when in the matrix there are nonzero elements  $|c_i, c_{i,j}| > |\pm \delta|$ , the values of which exceed the value of allowable

deflections of the values being compared for the unknown mixture being identified, from the corresponding values for the similar values of a standard sample of a known mixture;

**Example 3**

[0045] This example differs from Example 1 as follows: a step is added to measure intensity values of Raman scattering of a sample being identified and a standard sample, for which purpose the following steps are to be added:

[0046] a. Samples of an unknown mixture and a known mixture are irradiated, by turns, with a monochromatic line of a narrow-band (lined) source of light at one or several selected fixed frequencies  $\nu_r$ ;

[0047] b. The light of Raman scattering is gathered onto a photo-detector device;

[0048] c. For each of the extracted frequencies  $\nu_1, \nu_2, \dots, \nu_p$  of the selected spectral range, intensity values of the light gathered onto a photo-detector of the line are measured, namely: the exciting light falling on a sample  $I_{ex}$ , and the light of Raman scattering of a sample  $I_R$ , including steps of decomposition of the light of Raman scattering into a variety of spectral sections, and/or sequential extraction of the specified frequency sections  $\nu_1, \nu_2, \dots, \nu_p$  with a width of  $\Delta_r$ , with assigned pitch  $\Delta\nu_r \geq \Delta_r$ , where each section corresponds to the predetermined characteristic frequency of the light in the

selected spectral range  $\nu_r$ , which is gathered and registered by a photo-detector;

[0049] d. The relative intensity of the gathered light of Raman scattering at each of the extracted frequencies  $\nu_1, \nu_2, \dots, \nu_p$  for a known and unknown mixtures is defined in accordance with the following expressions:

$$R^K(\nu_{ex}, \nu_p) = \frac{I_R^K(\nu_{ex}, \nu_p)}{I_{ex}^K}; \quad R^U(\nu_{ex}, \nu_p) = \frac{I_R^U(\nu_{ex}, \nu_p)}{I_{ex}^U};$$

where:

$I_R^K(\nu_{ex}, \nu_p), I_R^U(\nu_{ex}, \nu_p)$  - intensity of Raman scattering irradiated by a sample of a known  $K$  mixture and a sample of an unknown  $U$  mixture within the specified sections  $\nu_1, \nu_2, \dots, \nu_p$  of the extracted frequency range  $\nu_r$  with light excitation at one of the selected frequencies  $\nu_{ex}$ ;

$I_{ex}^K, I_{ex}^U$  - intensity of the exciting light falling on corresponding samples at the time of performing measurements;

[0050] f. Data for an unknown mixture being tested with are compared with similar data for a standard sample of a known mixture by the following expression:

$$A = \frac{1}{2} \times \left( \frac{R^U(\nu_{ex}, \nu_p)}{R^K(\nu_{ex}, \nu_p)} \right) \times \left( \frac{1}{m} \times \sum_{m=1}^m \frac{T^U(\lambda_m)}{T^K(\lambda_m)} + \frac{1}{m \times n} \times \sum_{m=1}^m \sum_{n=1}^n \frac{L^U(\lambda_m, \lambda_n)}{L^K(\lambda_m, \lambda_n)} \right),$$

at the same time, a conclusion on the identity of the unknown mixture and the known mixture may be drawn when  $A = 1 \pm \delta$ ;

#### Example 4

[0051] This method differs from that of Example 2 as follows: comparison is performed for electronic signatures containing spectral profiles of absorption, luminescence and Raman scattering intensities for an unknown mixture sample ( $RALS_U$ ) and a standard sample ( $RALS_K$ ), which are formed by the method of Example 3, resulting in the following step of normalization of Raman matrixes in the following way:

[0052] a. A calibration sample  $Q_R$  is prepared, which calibration sample represents a specially made-up mixture or a multi-component solution of a strictly identical constant compound, the luminescence spectrum of which overlaps with the Raman spectrum of the known mixture within the selected analytical spectral ranges;

[0053] b. The intensity of Raman scattering is measured, and the results are converted similarly to steps b-f of Example 2. The signatures of a standard sample and a sample being identified are formed in accordance with the following expressions:

$$\|K_R\| = \left\| \begin{pmatrix} \frac{R_{1,1}^K}{L_{1,1}^{Q_R}} \dots \left( \frac{R_{1,p}^K}{L_{1,p}^{Q_R}} \right) \\ \dots \\ \left( \frac{R_{q,1}^K}{L_{q,1}^{Q_R}} \right) \dots \left( \frac{R_{q,p}^K}{L_{q,p}^{Q_R}} \right) \end{pmatrix} \right\| \quad \text{и} \quad \|U_R\| = \left\| \begin{pmatrix} \frac{R_{1,1}^U}{L_{1,1}^{Q_R}} \dots \left( \frac{R_{1,p}^U}{L_{1,p}^{Q_R}} \right) \\ \dots \\ \left( \frac{R_{q,1}^U}{L_{q,1}^{Q_R}} \right) \dots \left( \frac{R_{q,p}^U}{L_{q,p}^{Q_R}} \right) \end{pmatrix} \right\|, \quad \text{where}$$

$R_{q,p}^K, R_{q,p}^U$  - intensity values of Raman light scattering, normal-

ized for intensity of the exciting light  $I_{ex}$ , for samples of the known  $K$  mixture and the unknown  $U$  mixture, measured at the frequencies  $I_p$  with light excitation at the frequency  $I_q$ ;

$L_{q,p}^{Q_R}$  - the luminescence intensity for a calibration sample  $Q_R$ , normalized for the intensity of the exciting light  $I_{ex}$ ;

[0054] c. The electronic signatures of an unknown mixture  $\|U_E\|$  being tested are compared with similar data for a standard sample of a known mixture  $\|K_E\|$ , including the step of comparing Raman matrixes  $(RALS_{U,K})$ , by the following expression:

$$\|A_{RALS}\| = \left( \frac{\|U_R\|}{\|K_R\|} \right) \times \left( \frac{\|U_E\|}{\|K_E\|} \right),$$

where the identity of samples is defined by the following attribute:  $\|A_{RALS}\| = 1 \pm \delta$ ;

#### **Example 5**

[0055] The process of this embodiment differs from the processes of Examples 1 and 2 as follows: a step is added to dissolve samples of a mixture being identified and a certain standard mixture in appropriate solvents.

[0056] The most effective realization of the complex analysis system of the present invention is performed by a specialized multifunctional measuring device which is able to perform relative measurements of absorption spectra and luminescence, as well as Raman scattering, for samples under comparison in a



common analytic cycle. This makes it possible to minimize both systematic and random inaccuracies of measurements, and provides a high degree of reliability.

[0057] The apparatus of the present invention is illustrated in block diagram in Figure 3 a. This diagram includes all of the functions of the analytical steps required for the analysis of the present invention.

[0058] To perform complex measurements of absorption spectra, luminescence and Raman scattering, a combined light source is used consisting of continuous and line light sources LS, and a matching device, thereby ensuring the possibility of switching and focusing the light from the required source onto a sample.

[0059] Step A of Example 1 is performed by a  $\lambda_1$ SM-module in the aggregate with a light source LS and a light-gathering device  $F_1$ ;

[0060] - a samples holder SH performs alternate irradiation of samples (Example 1, step b) and measurements of intensity values for the probing light  $I_0$ , the passing light  $I_T$ , and the luminescent light  $I_L$  of a standard sample K and an analyzed sample U (Example 1, step d), which are performed with different positions of the holder in regard to the analyzing light beam (Fig. 3 b-d);

[0061] - gathering the light falling on a sample and passing through it is performed by a light gathering device  $F_2$  to a photo-detector  $PD_1$ , and gathering the luminescent light is performed by a light-gathering device  $F_3$  to a photo-detector  $PD_2$ , at the same time the luminescence light is disintegrated into spectral components, and the required spectral section is extracted from this light by a  $\lambda_j$ SM-module (Example 1, step c);

[0062] - defining the relative intensities of the passed light and the luminescent light (Example 1, step e), which represent experimentally measured intensity values for the passed light  $I_T$  and the luminescent light  $I_L$  normalized for  $I_0$ . This is performed while processing signals are measured at different positions of the sample holder SH (Fig. 3 b-d), by the devices included in a control and registration module (R&RM), and/or further mathematical treatment implemented by an external computing device;

[0063] - the comparison of corresponding relative intensities (Example 1, step f) is performed similarly to ascertain the identity or difference of samples being analyzed;

[0064] - the R&RM module also contains devices to control measurements ensuring switching of sample positions, setting the specified intervals of the extracted wavelengths, and measuring signals with a specified time constant, as well as power units of light sources and photo-detectors (PMT) ensur-

ing stabilization and automatic correction of power supply modes, and an interface of communication with an external computing device and/or devices of accumulation, storage and display of information.

[0065] The algorithm of comparison of characteristics of a known sample and an unknown sample provided in the present invention gives an unambiguous characteristic for the mixture being identified, whether it corresponds with a standard sample or not. Inconsistency between a sample being identified and a standard sample is evidence of a difference in their component compounds. The compliance level for a mixture being identified and a standard sample may be characterized by the value of correlation of their absorption-luminescent signatures  $\Theta$ . In the simplest case, this correlation is expressed by the ratio of a number of coincident elements of matrixes  $n_{i,j}$ , in accordance with the analytical expressions of the methods by claims 1-4, and the total number of significant elements of matrixes  $N$ :

$$\Theta = n_{i,j} / N;$$

This is illustrated with the following nonlimiting examples:

### **Example 6**

[0066] As an example of liquids identification, the following liquids are chosen (Table 1):

- two samples of drinking water from different sources;
- two samples of food spirit from different manufacturers;
- two samples of vodka from different manufacturers;
- two samples of motor gasoline of different types;
- two samples of shampoo of the same type from different manufacturers.

Each of the above-listed objects, by turns, is considered as a standard sample and compared by turns with the whole aggregate of tested samples for the compliance level in accordance with the method of Examples 1 and 2. As one can see in Table 1, compliance is not observed for a standard sample and tested ones for all comparisons, with the exception of total identity of samples. At the same time, objects having the same micro-compound elements (water, ethanol, vodka) have the higher compliance level.

Table 1.

Control sample		Compliance level									
Liquid type	№	1	2	3	4	5	6	7	8	9	10
Water 1	1	1	0.59	0.7	0.67	0.67	0.56	0.35	0.3	0.2	0.15
Water 2	2	0.59	1	0.68	0.66	0.63	0.52	0.42	0.37	0.25	0.28
spirit 1	3	0.7	0.68	1	0.9	0.77	0.64	0.15	0.1	0.11	0.15
spirit 2	4	0.67	0.66	0.9	1	0.76	0.61	0.12	0.1	0.15	0.12
Vodka 1	5	0.67	0.63	0.77	0.76	1	0.6	0.25	0.2	0.23	0.18
Vodka 2	6	0.56	0.52	0.64	0.61	0.6	1	0.23	0.18	0.25	0.28
gasoline 1	7	0.35	0.42	0.15	0.12	0.25	0.23	1	0.68	0.42	0.37
gasoline 2	8	0.3	0.37	0.1	0.1	0.2	0.18	0.68	1	0.35	0.32
Shampoo 1	9	0.2	0.25	0.11	0.15	0.23	0.25	0.42	0.35	1	0.4
Shampoo 2	10	0.15	0.28	0.15	0.12	0.18	0.28	0.37	0.32	0.4	1

[0067] It should be noted that ascertaining the identity of samples by comparing their differences, within the framework of the present invention, presents different tasks which differ in the amount of time required. To ascertain unambiguously the identity of objects being compared, it is necessary to determine the compliance by the greatest possible number of

parameters, whereas, for ascertaining their difference, it is enough when at least one of the parameters under comparison for a mixture being identified differs from a corresponding parameter for a standard sample. This is illustrated by Examples 7 and 8.

#### **Example 7**

[0068] In Table 2, a comparison of compliance levels is presented for pure liquids which were obtained from correlating absorption-luminescent signatures (ALS) in accordance with the method shown in Examples 1 and 2, and the same signatures supplemented by the Raman co-factor (RALS) in accordance with the processes of Examples 3 and 4. As can be seen in Table 2, adding the Raman component does not substantially change the value of the conformity level for objects related by being in the same chemical class, and results in drastic differences for objects which are in different chemical classes.

**Table 2.**

Identity method	Control sample	water		spirit		cyclohexane
		1	2	3	4	5
ALS	1	1	0.59	0.7	0.67	0.25
	2	0.59	1	0.68	0.66	0.2
	3	0.7	0.68	1	0.9	0.4
	4	0.67	0.66	0.9	1	0.35
	5	0.25	0.2	0.4	0.35	1
RALS	1	1	0.59	0.18	0.17	0
	2	0.59	1	0.17	0.16	0
	3	0.18	0.17	1	0.85	0,3
	4	0.17	0.16	0.85	1	0.26
	5	0	0	0.3	0.26	1

### **Example 8**

[0069] In Figure 4, correlation spectra are shown for intensity values of impurity luminescence using the same fixed excitation wavelength (230 nm) for water taken from various points of the municipal water supply system (curves 1-3 on Fig. 4) and for the same water contaminated with municipal wastewater (curve 4); 3,4-benzopyrene (curve 5); and petroleum derivatives (curve 6). As one can see from Figure 4, all three samples of clean water have minimal differences, while at the same time the presented samples have perceptible differences using the complete set of parameters (this value for samples 1, 2, and 3, respectively, are 0.86, 0.9 and 0.95). It is clear that contaminants in the water change the situation drastically, so that users can determine that these are distinct from a standard sample only on the basis of the date presented, which reduces the analysis time by a factor of about 10-20.

### **Example 9**

[0070] This example shows identification of a variety of industrial products produced for domestic purposes:

[0071] Three samples of dry yeast, two of which belong to the same production lot of the same manufacturer (samples 1-1 and 1-2), and a sample of a similar product from another manufacturer (sample 2);

[0072] Three samples of washing powder of the same brand from the same production lot from a well-known manufacturer (samples 1-1 and 1-2), and a sample of a similar product of another brand from the same manufacturer (sample 2);

[0073] Three samples of domestic gel-like washing liquid of the same brand, from the same production lot, from a well-known manufacturer (samples 1-1 and 1-2), and a sample of a similar product from the same manufacturer, but another brand (sample 2);

[0074] One can distinctly see in Table 3 that samples of products manufactured from strictly identical raw materials during the same processing cycle have a high compliance level with each other. This compliance is not observed when the manufacturing conditions are changed.



**Table 3.**

	№	dry yeast			washing powder			domestic gel-like washing-up liquid		
		1-1	1-2	2	1-1	1-2	2	1-1	1-2	2
dry yeast	1-1	1	0.95	0.5	-	-	-	-	-	-
	1-2	0.95	1	0.65	-	-	-	-	-	-
	2	0.5	0.65	1	-	-	-	-	-	-
washing powder	1-1	-	-	-	1	0.86	0.3	-	-	-
	1-2	-	-	-	0.86	1	0.4	-	-	-
	2	-	-	-	0.3	0.4	1	-	-	-
domestic gel-like washing-up liquid	1-1	-	-	-	-	-	-	1	0.9	0.45
	1-2	-	-	-	-	-	-	0.9	1	0.4
	2	-	-	-	-	-	-	0.45	0.4	1

**Example 10**

[0075] This example illustrates determining the impurities contaminating liquids, in this case, drinking water, with additives of different classes of substances. As a standard sample, clean drinking water was accepted here. The following liquids were tested for the presence of contaminants:

The standard water

The standard water with priority PAH ( 3,4-benzopyrene)dissolved therein;

The standard water with crude petroleum dissolved therein;

The same water with organophosphorus insecticides dissolved therein : Thiophos (structure 1) and foksim (structure 2);

The standard water with chlorine dissolved therein.

The standard water with medical products dissolved therein: methyl ether of benozylecognine (structure 3); 1,3,7-trimethylxanthine (structure 4); penicillin (structure 5); and tetracycline (structure 6);

The same water containing *E. coli*;

The same water contaminated with municipal wastewater.

[0076] This example demonstrates the adaptability of the method of the present invention in monitoring water systems for the purpose of exposing contamination with either chemical or biological contaminants.

[0077] As can readily be seen from Table 4, the compliance level of water contaminated with 3,4-benzopyrene with eth standard water was reduced to one half, which compliance was reduced even more when the water was contaminated with petroleum. A similar situation was also observed in the case of

contamination with substances such as organophosphorus insecticides, chlorine, medical products, bacteria, and municipal wastewater.

[0078] It should be noted that adding the chlorine resulted in a drastic reduction of the conformity level of the ALS signature of a sample being tested with a sample of clean water. At the same time, visual checks of outline maps of these objects (see Figure 2) do not permit the user to draw the same conclusions. These facts point out the high level of accuracy of the present invention.

**Table 4.**

Contaminant	Compliance level		
	Is absent	3,4-benzopyrene	Petroleum
PAH	0.5±0.05	0.48	0.39
	Is absent	Structure 1	Structure 2
Organophosphorus compound	0.95±0.05	0.43	0.41
	Is absent	0,41	
Chlorine	0.95±0.05		
	Is absent	Structure 3	Structure 4
Medical products	0.95±0.05	0.32	0.46
	Is absent	Structure 5	Structure 6
	0.95±0.05	0.4	0.36
	Is absent	e-coli	
Bacteria culture	0.95±0.05	0.5	
	Is absent	0.55	
Municipal wastewater	0.95±0.05		
	Is absent		

[0079] The foregoing description of the specific embodiments will so fully reveal the general nature of the invention that others can, by applying current knowledge, readily modify

and/or adapt for various application such specific embodiments without undue experimentation and without departing from the generic concept. Therefore, such adaptations and modifications should and are intended to be comprehended within the meaning and range of equivalents of the disclosed embodiments.

[0080] It is to be understood that the phraseology or terminology employed herein is for the purpose of description and not of limitation. The means and materials for carrying out various disclosed functions may take a variety of alternative forms without departing from the invention.

[0081] Thus, the expressions "means to..." and "means for..." as may be found in the specification above and/or in the claims below, followed by a functional statement, are intended to define and cover whatever structural, physical, chemical, or electrical element or structures which may now or in the future exist for carrying out the recited function, whether or not precisely equivalent to the embodiment or embodiments disclosed in the specification above. It is intended that such expressions be given their broadest interpretation.

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